UNIVERSITY OF GONDAR

FACULTY OF VETERINARY MEDICINE

PREVALENCE OF GASTRO INTESTINAL NEMATODE PARASITIC INFECTIONS OF HORSES AND DONKEYS IN AND AROUND KOMBOLCHA TOWN

DVM THESIS

BY

SAMUEL ENGDAW MOLLA

JUNE, 2015

GONDAR, ETHIOPIA

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A thesis submitted to the Faculty of Veterinary Medicine, University of Gondar in partial fulfillment of the requirements for the degree of Doctor of Veterinary Medicine

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LISTS OF ABBREVIATIONS

BCS	Body conditions
CSA	Central statistical authority
EPG	Eggs per gram
GIT	Gastro-intestinal tract
^{O}C	Degree cent grade
MIDROC	Mohammed International Development Research and Organization Companies
SPSS	Statistical package for social science
SWZARDO	South Wollo Zone Agricultural and Rural Development Office
UCI	Upper central incisors

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ABSTRACTS

A cross sectional study was conducted from November 2014 to April 2015 in and around Kombolcha town to know the prevalence and the types of gastrointestinal nematode parasites of donkeys and horses. A total of 384 donkeys and horses (253 donkeys and 131 horses) were randomly selected and examined during the study period. During the examination, floatation technique was used and the overall prevalence of gastrointestinal nematode parasites was 76.04% (292 from 384) of which 86.5 % is in donkeys and 55.7% in horses. The prevalence of *Strongyle*, *Parascaris equorum* and *Oxyuris equi* was 47.4%, 4.7% and 2.8% in donkeys and 36.6%, 6.9% and 3.1% in horses respectively. There were also mixed infestations like *Strongyle* and *Parascaris equarum, strongyle* and *Oxyuris equi, strongyle, Parascaris equi* and *Oxyuris equi* with total prevalence of 12.5%, 5.5%, and 6% respectively. The risk factors, species, sex, age, body conditions and origins having respective p-values of 0.000, 0.037, 0.028, 0.009 and 0.000 were significantly associated with the occurrence of GIT Nematode parasites. Donkeys were found more likely at risk of developing gastrointestinal nematode parasites than horses (OR = 5.1, 95% CI = 3.1- 8.4).The age related prevalence was 85.1%, 71.77%, 80% respectively for young, adult and old ages while 70.2%, 81.3% and 64.4% prevalence was for poor, medium and good body mass conditions respectively. In conclusion, the current study revealed higher prevalence of gastrointestinal Nematode parasites of horses and donkeys in the area. Therefore, regular deworming, improvement of housing and feeding management systems and awareness creation to animal owners were recommended.

Key words: *Donkeys, GIT Nematode parasites, Horse, Kombolcha, Prevalence, Risk factors*

1. INTRODUCTION

In the developing world, there are estimated 110 millions of equines. Ethiopia has a large numbers of equines. It has a total of 9.83 millions equine populations. From those numbers, donkeys accounts 7.04 million while horses and mules are 2.03 and 0.4 million respectively (CSA, 2014/15). Equines have a prominent position in the agricultural systems of many developing countries. In Ethiopia, the low level of development of the road transport network and the rough terrain of the country make the donkeys and the horses the most valuable, appropriate and affordable pack animals under the small holder farming system (Hailu *et al.*, 2013).). In Kombolcha, which is found in south wollo zone administration, there are both horses and donkeys. Horses in the area are all cart horses in the town while donkeys are found around the town.

Equines play an important role as working animals in many parts of the world, employed for packing, riding, carting and ploughing. Equine power is vital for both rural and urban transport system which is cheap and provides the best alternatives in places where the road network is insufficiently developed and in the cities where narrow streets prevent easy delivery of merchandise. It is suggested that donkeys can play a great role in the frame works of food security and social equity of high food insecure countries (Regassa *et al*., 2013).

In areas away from roads, many people use mules and donkeys to transport food and other supplies to villages. Long working hours and difficult conditions are experienced by donkeys and mules. These animals are often engaged in work for long hours and when get free, they are left to browse and feed on garbage. These have the potential to affect negatively on their welfare of life and health (Bogale, 2012.

Parasitic helminthes are one of the most common factors that constrain the health and working performance of donkeys and horses world widely. They cause various degrees of damage depending on the species, nutritional and the immune status of equines. They decrease the performance and productivity in the animals mainly, in the reduction of body weight or failure to gain weight or even increase the mortality in acute case (Mezgebu, 2013).

Equines are hosts to great nematodes of the family *Strongylidae*, commonly called *Strongyle* nematodes or *Strongyles*. The most common gastro intestinal nematode parasites of equines include large *strongyles*, small *strongyle*, *Ascaris* and pinworms (*Oxyuris equi)* (Woruku and Afera, 2012). Some studies were conducted in the town in horses. But, this current study includes both donkeys and horses.

The Objectives of the study are:

- To know the current prevalence of gastrointestinal nematode infections of horses and donkeys in and around Kombolcha town.
- To identify the types of gastrointestinal nematode parasites of donkeys and horses in the study area.

2. LITERATURE REVIEW

Nematodes in general

Nematodes are round worms make up a large assemblage of relatively simple structure with a wide spread distribution. Their cylindrical, non segmented bodies distinguish them from other helminthes (Mandel, 2006). Nematodes do not have circular muscle layer, rather all of circular musculatures are oriented longitudinally and divided into dorsal and ventral fields (Bowman, 2003). Most of the free living nematodes are microscopic as many of the parasitic species invade the body fluids such as the blood or lymph channel of their hosts (Ballweber, 2001).

Nematodes have separate sex and male nematodes are smaller than females of their species (Bowman, 2003). The horse is host to a great number of gastrointestinal parasite species. From such a parasitic infection of nematodes, some important are: Strongylosis, Oxyridosis, Strongyloides (thread worms) and Ascaridosis (Mandel, 2006).

2.1. Strongylosis

2.1.1. Etiology

The red worms (strongyles) are nematodes commonly found in the large intestine of horses and other equids. They belong to two sub families: the Strongylinae (large stronglyes) and Cyathostominae (known variously as small *strongyles,* small red worms, *trichonemes, cyathostomes* or *cyathostomins*) (Radostits *et al*., 2007).The large *strongyles* include *S.vulgaris, S. edentatus, and S. equinus*, which migrate extensively through the body, and *Triodontophorus* spp. and *Oesophagodontusrobustus,* which do not. The *cyathostomins* comprise more than 40 non migratory species. Of these, about 10 species occur commonly (Ballweber, 2001).

2.1.2. Epidemiology

It is distributed world widely and most common in grazing equids. Eggs are shed by horses of all ages. Infective larvae develop seasonally on pasture. Hypobiotic cyathostomin larvae can cause severe disease when they resume development in late winter (Kassai, 1999).

2.1.3. Clinical signs

Ill-thrift, weight loss, poor hair coat, and impaired performance, Verminous arthritis (associated with *Strongylus vulgaris*) variable, including colic and diarrhea. Larval cyathostominosis cause rapid weight loss on clinical examination. The mucosa is pale, the heart rapid and loud and respiration moderately increased. Intestinal sounds are increased although the feces are normal. Abortion may occur and the mare usually dies (Ballweber, 2001). `

2.1.4. Lifecycle

The life cycle of all species is direct. Eggs are passed in the feces and under suitable climatic conditions produce infective third-stage larvae from 7 days onwards. Horses become infected by ingestion of the infective larvae (Kassai, 1999). After ingestion, the larvae of non migratory *strongyles*, such as the *cyathostomins*, exsheath and enter the walls of the cecum and colon before breaking out into the lumen of the intestine. Larvae of *S. edentatus* penetrate the intestine and travel via the portal vessels to the liver, where larvae remain and produce hemorrhagic tracts for a month. They then migrate via the hepatho renal ligament to the connective tissue. After about 3 months, they return via the root of the mesentery to the large bowel wall and again form hemorrhagic nodules, which finally rupture and release the worms into the lumen (Taylor *et al*., 2007).

Larvae may be found in other organs, e.g. the testes, but these larvae do not return to the intestine. *S. equinus* migrates via the liver to the pancreas and peritoneal cavity but how they return to the intestine is uncertain. Larvae of *S. vulgaris* penetrate the intestinal wall, molt to the fourth larval stage in the sub mucosa and then pass into and up small arteries. Nodules are formed in the intestine wall and later rupture, releasing adults into the lumen of the intestine. The prepatent period is 6 months (Radostitis *et al*., 2007).

2.1.5. Pathogenesis

The disease processes associated with the *strongyles* can be divided into those produced by migrating larvae, those provoked by the mass emergence of mucosal larvae and those associated with adult worms. Heavy intestinal infection can alter intestinal motility, permeability, and absorption. The larvae of *S. vulgaris* are the most pathogenic, causing arthritis, thrombosis and thickening of the wall of the cranial mesenteric artery. Emboli may

break away and lodge in smaller blood vessels, leading to partial or complete ischemia in part of the intestine, thus producing colic. Colic may also be caused by pressure of the thickened cranial mesenteric artery on the mesenteric plexus (Kassai, 1999).

2.1.6. Necropsy findings

Necropsy findings usually include most of the lesions characteristic of each worm. In cases of general strongylosis, very large numbers of adult worms will be found in the cecum and colon. Adult *strongyle* worms may be seen attached to the mucosal surface. The three Strongylus spp. are red in color and 2-5 cm long. *Triodontophorus* and *Oesophagodontus* are smaller, up to 2 cm. Catarrhal, hemorrhagic or fibrinous inflammation of the cecum and ventral colon with multiple small ulcers is associated with the emergence of *cyathostomin* larvae (Radostitis *et al*., 2007).

2.1.7. Diagnostic confirmation

A specific diagnosis is difficult to achieve in every case. Few clinical observations or laboratory results are pathognomonic for the disease syndromes associated with *strongyle* infection (Lefevre, 2010).

2.1.8. Treatment

Treatment may be targeted against immature and adult large and small strongyle worms in the lumen of the intestine. Anthelmintics vary in their efficacy against these larval stages. For elimination of adult worms there is a wide choice of compounds and formulations for use in feed.Ivermectine:0.2mg/kg,moxidectin:0.4mg/kg,.Benzimidazoles(febantel6mg/kg,fenbendazole7.5mg/kg,mebendazole5-10mg/kg,oxibendazole10mg/kg),Tetrahydropyrimidines(6.6mg pyrantel base/kg) (Taylor *et al*., 2007).

2.1.9. Control

Control should not rely solely on anthihelmentics. Judicious uses of drugs of key times of the year combined with improved pasture management are key ingredients of control (Foryet, 2001).

2.2. Strongyloides

Farm animals in many countries are exposed to infection with the nematode genus Strongyloides. Disease outbreaks occur in young pigs and foals. But the overall economic importance of this parasite does not appear to be very great. Different species occur in each host: *Strongyloides ransomi* in pigs, *S. westeri* in horses. All are parasites of the small intestine. They are thread-like and less than 1 cm in length. Only female worms are present in the intestine and so eggs are produced by parthenogenesis. The eggs are thin shelled and contain an embryo. The larvae that hatch out may develop into infective or non-parasitic forms (Mandel, 2006).

2.2.1. Transmission

It occurs when the infective larvae enter in to the host either by ingestion or by skin penetration. In older animals, they accumulate in subcutaneous tissues and migrate to the mammary gland when lactation starts. Neonates are thereby infected via the milk (Lefevre, 2010).

2.2.2. The life cycle

It is direct life cycle. The eggs are very resistant to external environmental conditions. An infective larva develops inside the egg but a relatively high temperature is required for rapid growth.

2.2.3. Clinical signs:

Diarrhea in young animals is the most common but the passage of massive numbers of larvae through the skin may also provoke dermatitis. In foals, high egg counts may be recorded in apparently healthy animals. But, it may coincide with the onset of diarrhea (independent of the first heat of the mare) in other individuals (Ballweber, 2001).

2.2.4. Pathogenesis

Infection in older animals is apparent and usually so in young animals. Disease when it occurs is present only in young animals (Ballweber, 2001).

2.2.5. Diagnosis

It depends on detection in the feces of the yellow oval eggs, which have a transparent plug at each end. The eggs are heavier than many others and do not always float well in saturated salt sodium chloride solution. An alternative flotation fluid such as zinc sulfate or sugar is more reliable. At necropsy, the adult worms which are 2-5 cm long are easily recognized by their whip-like appearance- the anterior third is much thinner than the handle-like posterior end (Foryet, 2001).

2.2.6. Treatment

Most broad spectrum anthelmintics are effective in eliminating this parasite. In foals, ivermectin is used at the standard equine dose. But, elevated doses of fenbendazole (50 mg/kg) and oxibendazole (15 mg/kg) are needed. The treatment of mares with ivermectin on the day of parturition did not prevent trans mammary transmission but markedly reduced egg counts in the foals (Radostitis *et al*., 2010).

2.2.7. Control

Control depends on the elimination of warm, moist areas such as damp litter or bedding suitable for parasite multiplication (Mandel, 2006).

2.3. Oxyridosis

2.3.1. Etiology: *Oxyuris equi*

2.3.2. Clinical signs

Oxyuris equi is a nematode that provokes irritation of the perennial region of horses, causing them to rub and bite their tails. This can result in hair loss and sometimes physical damage to the tissues of the area. The parasite is ubiquitous but of greater prevalence in areas of high rainfall (Radostitis *et al*., 2007).

2.3.3. Life cycle

It is a direct life cycle. Adult females migrate to the rectum out anus and cement masses of eggs on perineal area, infective L3 develops within the egg in 4-5 days, environmental contamination occurs when cementy fluid dries and cracks allowing eggs to fall off or egg masses are rubbed off by horses. Infections by ingestion of eggs contain infective L3 hatche in

small intestine and larvae move to cecum where they mature. Prepatent period is 4-5 monthes. Transmission then occurs via contaminated feed stuffs (Ballweber, 2001).

2.3.4. Diagnosis

Diagnosis is by detection of operculated eggs, slightly flattened on one side, on transparent adhesive tape that has been pressed against the peri anal skin and then placed on a microscope slide for examination, or by the chance observation of an adult worm in the feces. The mature worms are gray in color and inhabit the cecum and colon. The male is 1-2 cm long, but the female is much longer, up to 15 cm, and has a long tapering tail (Urquart, 2003).

2.3.4. Treatment

It comprises the application of a mild disinfectant ointment to the peri anal region and the administration of ivermectin, moxidectin, any of the newer broad-spectrum benzimidazoles or pyrantel at the standard dose rate for horses. Piperazine salts are also effective (Ballweber, 2001).

2.3.5. Control

Wash every 4 days of the perineal area and underside of the tail head with soap and water to remove egg masses (Urquart, 2003).

2.4. Equine parascariosis

2.4.1. Etiology

It is aused by *Parascaris equorum*. It is one of the largest nematode species ranging 20-50 cm in length. It is cosmopolitan (Bowmann, 2003).

2.4.2. Epidemiology and Distribution

It has worldwide distribution with more prevalent in grazing equines. It is highly pathogenic in foals. In horses, the only route of infection is by ingestion of larvated eggs. Because the eggs have very thick walls, the infective stage is protected from deleterious environmental influences. Few disinfectants will harm them and they are very resistant to cold. But survive most readily in cool, moist surroundings. Eggs are seen occasionally in the feces of very young foals. But, this is thought to be due to the ingestion of uninfective eggs during coprophagia (Kassai, 1999).

2.4.3. Lifecycle

The adult worms live in the small intestine and lay very large numbers of thick shelled eggs. These are not infective until a larva has developed inside. This process needs suitable warmth and humidity and takes place over a period of several weeks. When swallowed, infective eggs hatch quickly in the intestine of the host and the larvae migrate through the intestinal wall, reach the portal vein and are transported to the liver. They cross to the hepatic venous system and travel to the lungs, are passed up the bronchi and trachea to the pharynx, are swallowed and come to rest in the intestine where they mature (Taylor et al., 2007). The prepatent period (time from infestation to the appearance of eggs in the feces) is 11-15 weeks.

2.4.4. Pathogenesis

The Migrations of the larvae through the liver cause hemorrhage and fibrosis. The most serious damage occurs in the lungs where the larvae provoke alveolar injury with edema and consolidation. Foals with *P. equorum* have reduced gut motility, an increase in the ratio of body water to body solids and a lowering of the body pool of albumin (Lefevre, 2010).

2.4.5. Clinical findings

Clinical signs include frequent coughing, bilateral mucoprulent discharge, decreased feed intake, un thriftiness, colic, diarrhea, and death (Radostitis *et al*., 2007).

2.4.6. Necropsy findings

In the early stages of a massive infestation, there are sub pleural hemorrhages, and edema and congestion of the lungs. The pleural cavity may contain blood-stained fluid. The liver is enlarged and congested and there may be hemorrhages under the capsule. Microscopically, necrotic tracts and sections of larvae are observed alveolar wall thickening with fibrin (Radostitis et al., 2007).

2.4.7. Diagnostic confirmation

Ascarid eggs are brown and have thick walls with a pitted surface. Fecal egg counts in excess of 1000 epg are considered to be indicative of significant infection. Migrating larvae are too small to be observed by the naked eye at postmortem examination. They can be recovered from macerated lung tissue by the Baermann technique or seen microscopically in scrapings of bronchial mucus (Ballweber, 2001).

Differential diagnosis

Early stages of massive infection: Chronic form of *Rhodococcus equi* pneumonia in young foals, other causes of un thriftiness including malnutrition and chronic enteritis due to infections with *Salmonella* and *Brachyspina* species (Radostitis *et al*., 2007).

2.4.8. Treatment

In horses, ivermectin 0.2 mg/kg, moxidectin0.4 mg/kg, febante l6mg/kg, fenbendazole7.5 mg/kg, mebendazole5-10 mg/kg, oxibendazole10 mg/kg, and pyrantele mbonate19 mg/kg are all effective against adult *P equorum*. Ivermectin, moxidectin,and fenbendazole are also active against immature forms in the intestine (Taylor *et al.*, 2007).

2.4.9. Control

Young animals are most susceptible. Emphasis must be placed on preventing the environment from becoming contaminated. This is achieved by periodic treatment of the animals likely to be shedding eggs asymptomatic adult carriers as well as the more vulnerable young stock. Exposure of young foals to contaminated soil or bedding should be avoided (Andersen, 2000).

3. MATERIALS AND METHODS

3.1. Study area

A cross sectional study was was conducted from November, 2014 to April, 2015 in and around Kombolcha town, Ethiopia. Kombolcha is acity and woreda in north-central Ethiopia, in Amhara National Regional State, South Wello Zone, at a distance of 13 km to the north west of Dessie, 377 Km from Addis Ababa, and 505 Km from Bahirdar. It has a latitude and longitude of 11°5′N 39°44′E with an elevation between 1842 and 1915 meters above sea level with average annual temperature of 15.9 °C and average annual rainfall of 1248 mm. According to South Wollo Zone Agricultural and Rural Development Office (SWZARDO), there are 2540 horses, 634 mules, 7758 donkeys, and 1865 camels (SWZARDO, 2013).

3.2. Study population

The study was conducted on both horses and donkeys. These animals were found from different areas of Kombolcha towns and its surroundings. They were managed under extensive smallholder and semi-intensive husbandry systems. In the study area, equines were allowed to graze on communal or private owned pasture land. But, cart horses in the town feed on some supplementary feeds. The major feed resources in the area were natural pasture, hay, and crop residues. The animals were housed in houses with muddy grounds roofed with either hay or corrugated iron. Strategic de-worming was not practiced by the farmers of the area. But, cart horses in the town get occasional de-worming than around the town.

3.3. Study Design

A cross sectional study was conducted in randomly selected horses and donkeys for the detection of the prevalence of gastro intestinal Nematode parasitic infections. Informations about species, sex, age and body conditions of the study animals were gathered appropriately. The ages were determined using dentitions and owners' information. Animals were categorized as young (< 4 years) adult (4-9 years), Old (> 9 years) (Netsanet *et al*, 2014).The body conditions were classified according to Svendsen (2008) as poor, medium and good.

3.4. Sample size determination

The sample size required for this study was determined according to Thrusfield (2005). Since there was no previous work done in this study area, 50% prevalence was taken as expected prevalence for sample size determination of this study. The other determinants considered in sample size determination were 95% confidence interval and 5% desired absolute precision. Hence the sample size is estimated as

$$N = \frac{1.96^2[P_{exp}(1-Pexp)]}{d^2}$$

Where; N= required sample size

P_{exp}= expected prevalence of nematode parasites

d^2= desired absolute precision

1.96= the value of "z" at 95% level of confidence

d=5%=0.5

$$N = \frac{1.96^2\ 0.5(1-0.5}{(0.5)^2} = 384$$

Using the above formula, 384 animals of Horses and Donkeys were examined.

3.5. Study methodology

Random fecal samples were collected directly from the rectum of the study animals using disposable plastic gloves and placed into universal bottles. Each sample was labeled with necessary informations and immediately transported to Kombolcha Animal disease diagnosis Laboratory. Samples were kept in refrigerator at 4°C when immediate processing was not possible. But, it was processed within 48 hours. Some samples were held using10% formalin. Parasitological examination was done by direct smear and flotation techniques following the standard procedures for nematode parasites and examined microscopically (10× and 40×). Identification of the eggs was made based on the basis of their morphology.

3.6. Data collection

The data were collected by simple random method by taking horse and donkeys that originate from Kombolcha and its surroundings. The fresh fecal samples were collected from the rectum of the horses and donkeys.

3.7. Data management and Analysis

The collected data were coded and entered into Microsoft Excel spread sheet and Statistical analyses was performed using SPSS version 16 software packages. Descriptive analysis was made to know the chi square and prevalence described as percentages. Univariate and multivariate logistic regressions were used to know the significance and to calculate degree of association between risk factors and gastro intestinal nematode parasites.

4. RESULTS

4.1. The overall prevalence of GIT nematodes of donkeys and horses

384 fecal samples were taken from 131 horses and 253 donkeys and examined for the presence of different gastrointestinal nematode parasites. From the examined animals, 73 horses and 219 donkeys were positive with respective prevalence of 55.7% and 86.5 % for different GIT nematodes. The overall prevalence of GIT nematode in horses and donkeys in the study area was found to be 76.04 %.

Animals	Numbers of animals examined	Numbers of positive animals	Prevalence	x^2	P-value
Donkey	253	219	86.5 %		
horse	131	73	55.7%	45.04	P=0.000
Total	384	292	76.04%		

Table 1: Over all prevalence of gastro-intestinal nematode parasites in donkeys and horses.

4.2. Analysis of different risk factors:

The logistic regression analysis of the risk factors indicated the presence of strong association between the occurrence of GIT nematodes infections and among the species of animals, ages, body conditions, sexes and animal origins. Sex, age, species, body conditions and origins with their respective p-values of 0.037, 0.027, 0.000, .009, and 0.000 are all significantly associated with the risk of being infected with GIT nematodes. Donkeys were found 5.1 times more likely at risk of developing gastrointestinal nematode parasites than horses (OR = 5.1).

The risk of being infected for male is 1.65 times more likely than females (OR=1.65). Youngs are 1.4 times more likely infected than olds while adults are 0.63 times less likely infected than olds. Poor and medium body conditions respectively are1.3 and 2.4 times more likely infected than good body mass conditions. Donkeys and horses from the origins of Ancharo, Chefew, Dirma and Kombolcha cart stations are less likely infected from origin Ligo respectively by 0.43, 0.36, 0.29 & 0.079 times.

Risk factors	Numbers of Examined animals	Numbers of positives and prevalence	X^2	P-value	OR(95% CI)
Species			45.0	0.000	
Donkeys	253	219 (86.6%)			5.1(3.1-8.4)
Horses	131	79 (55.7%)			
Sex			4.40	0.037	
Males	236	188 (79.7%)			1.65(1.01-2.6)
Females	148	104 (70.2%)			
Age			7.37	0.028	
Youngs	101	86 (85.1%)			1.4(1.23-3.8)
Adults	248	178 (71.77%)			0.63(0.26-0.89
Olds	35	28 (80%)			
Body conditions			9.75	0.009	
Poor	94	66 (70.2%)			1.30(1.15-2.60)
Medium	231	188(81.3%)			2.41(1.29-4.5)
Good	59	38 (64.4%))			
Origins			48.30	0.000	
Ancharo	62	54 (87.1%)			0.43(0.10-0.75)
Chefew	74	63 (85.13%)			0.36(0.02-0.48)
Dirma	68	56 (82.35 %)			0.29(0.07-0.68)
Kombolcha cart stations	130	72 (55.38%)			0.07(0.02-0.09)
Ligo	50	47 (94%)			

Table 2: Analysis results of different risk factors

4.3. Relative proportions of each gastro intestinal Nematode parasites

During the laboratory examinations, different GIT Nematodes eggs were found. These were *strongyles, parascaris equorum Oxyuris equi* and mixed infections of *Strongyle + parascaris equi*, *strongyles + Oxyuris equi* and *strongyles + parascaris equi + Oxyuris equi.*

The highest percentage was recorded for *Strongyles*, followed by mixed infections of *Parascaris equorum + Strongyles* , *Strongyles + Parascaris equorum + Oxyuris equi*, *Parascaris equorum* , *Oxyuris equi + Strongyles* and *Oxyuris equi* with their respective prevalence of 168 (43.5%), 48 (12.5%), 23 (6%), 21 (5.5%), 21(5.5%), and 11(2.9%) (figure 1).

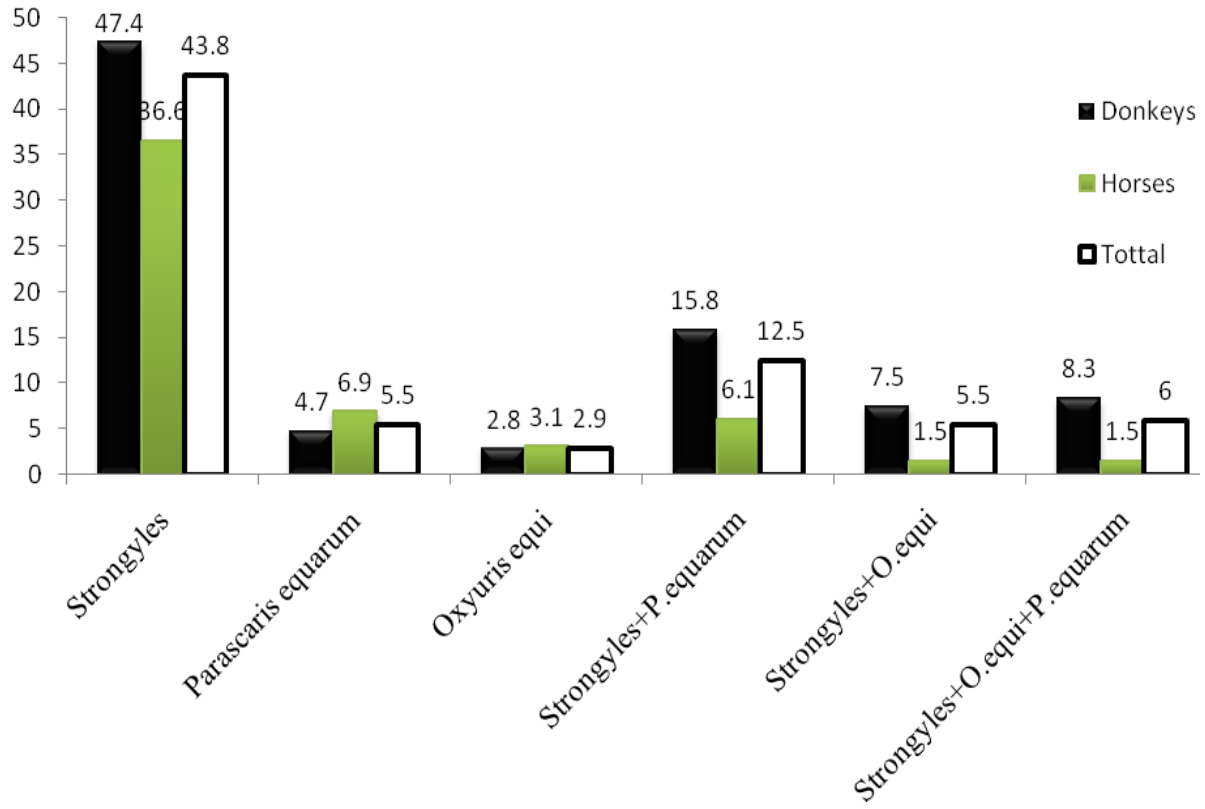

Figure 1: Proportions of each gastro-intestinal nematode parasite of donkeys and horses in %

4.4. Proportions of each gastro intestinal nematode parasite with risk factors

The highest prevalence of all the parasites was observed for *strongyles* infections in both species of donkeys (47.4%) and horses (36.6%) followed by mixed infections of *Strongyle* + *Oxyuris equi* (19 / 7.5% /) in donkeys and *Parascaris equarum* (9 / 6.9% /) in horses. *Strongyle* + *P. equarum* + *O. equi* (21 / 8.3% /), *Strongyle* + *Oxyuris equi* (40 / 15.8% /), *Parascaris equarum* (7 / 2.8 % /) for donkeys and *Strongyle* + *P.equorum*(8 / 6.1% /), *Oxyuris equi* (4 / 3.1% /), *Strongyle* + *Oxyuris equi* (2 / 1.5% /) *and Strongyle* +*Parascaris equarum*+*Oxyuris equi* (2 / 1.5% /) for horses held their respective ranks in each species.

Strongyles (105 / 44.5% /), *P.equorum*(14 / 5.9% /) and *Strongyle* + *Oxyuris equi* (16 / 6.8% /), have their highest prevalence in males while *O. equi* (5 / 3.4% /) and *Strongyle* + *P.*

equarum (20 / 13.5% /) are in females. *Strongyles* are high in prevalence in youngs (45 / 44.6% /) followed by adults (109 / 44% /) and olds (14 / 40% /), *Parascaris equarum* has highest prevalence in old (2 / 5.7% /) followed by adults (14 / 5.6% /) and youngs (14 / 5.6% /). *O. equi* accounts 5 / 5% /, 1 / 2.9 % / and 5 / 2 % / value respectively for youngs, olds and adults.

Strongyles found with highest prevalence in medium body conditions (117/ 50.6% /) followed by poor (35 / 37.2% /) and good BCS (16 / 27.1% /). *P. equarum* held high prevalence in poor body conditions (6 / 6.4% /) followed by medium (12 / 5.2% /) and good body mass (3 /5.1% /) while *O. equi* has prevalence of 4 / 6.8% /, 5 / 5.3 % /, 2 /0.9 % / respectively for good, poor and medium body mass conditions. *Strongyle* infections accounted highest prevalence for those around Ligo (29 / 58%) while *P. equoarum* in Dirma (5 / 7.4% /) and *O. equi* in Ligo (2 / 4 % /) (table 3).

Risk factors	Numbers of examined	*Strongyles*	*Parascaris equarum*	*Oxyuris equi*	*Strongyle + P. equarum*	*Strongyle + Oxyuris equi*	*Strongyle+P.equarum+Oxyuris equi*
		Positives / %/	Positives / % /	Positives /% /	Positives / % /	Positives / % /	Positives / % /
Species							
Donkeys	253	120 /47.4% /	12 / 4.7% /	7 / 2.8% /	40 / 15.8% /	19 / 7.5% /	21 / 8.3% /
Horses	131	48 / 36.6% /	9 / 6.9% /	4 / 3.1%	8 / 6.1% /	2 / 1.5%	2 / 1.5% /
Sex							
Female	148	63 / 42.6% /	7 / 4.7% /	5 / 3.4% /	20 / 13.5% /	5 / 3.4% /	4 / 2.7% /
Male	236	105/ 44.5% /	14 / 5.9% /	6 / 2.5% /	28 / 11.9% /	16 / 6.8% /	19 / 8.1% /
Ages							
Young	101	45 / 44.6% /	5 / 5% /	5 / 5% /	15 / 14.95% /	8 / 7.9% /	8 / 7.9% /
Adult	248	109 / 44% /	14 / 5.6% /	5 / 2% /	30 / 12.1% /	9 / 3.6% /	11 / 4.4% /
Old	35	14 / 40% /	2 / 5.7% /	1 / 2.9% /	3 / 8.6% /	4 / 11.4% /	4 / 11.4% /
Body conditions							
Poor	94	35 / 37.2% /	6 / 6.4% /	5 / 5.3% /	5 / 5.3% /	5 / 5.3% /	5 / 5.3% /
Medium	231	117/ 50.6% /	12 / 5.2% /	2 / 0.9% /	15 / 6.5% /	11 / 4.8% /	15 / 6.5% /
Good	59	16 / 27.1% /	3 / 5.1% /	4 / 6.8% /	3 / 5.1% /	5 / 8.5% /	3 / 5.1% /
Origins							
Ancharo	62	31 / 50% /	2 / 3.2 % /	1 / 1.6 % /	7 /11.3 % /	5 / 8.1 % /	8 /12.9 %/
Chefew	74	39 / 52.7 % /	5 / 6.8 %/	2 /2.7 %/	7 /9.5% /	5 / 6.8% /	5 / 6.8% /
Dirma	68	22 / 32.4% /	5 / 7.4% /	2 / 2.9% /	18 / 26.5% /	5 / 7.4% /	4 / 5.9% /
Kombolcha cart stations	130	47 / 36.2% /	9 / 6.9% /	4 / 3.1% /	8 / 6.2% /	2 / 1.5% /	2 / 1.5% /
Ligo	50	29 / 58%/	0	2 / 4% /	8 / 16% /	4 / 8% /	4 / 8% /

Table 3: Proportions of each gastro intestinal nematode parasites with each risk factor

5. DISCUSSIONS

In this current study, an overall of 76.04% prevalence of gastrointestinal nematode parasites with 55.7% in horses and 86.5 % in donkeys were obtained. This result (76.04%) agrees with the works of Tesfu *et al*. (2014) in Hawassa Town and Regassa and Yimer (2013) in South wollo zone with their respective results of 72.7% and 70.4%. This present finding is higher than the work of Sawsan *et al*. (2008) who reported with a prevalence of 29.79% of horse and 37.48% of donkeys in South Darfur state. But, it is relatively lower than reports of Mezgebu *et al.*(2013) , Ibrahim *et al*.(2011), and Ayele *et al.* (2006) with their respective results of 92.71% , 96.9%, and 98.2% for GIT helminthes parasite infection of equine at around Gondar, around Hawassa Town and Dugda Bora District, respectively.

This difference might be due to the differences in the study areas, deworming strategy and accessibility to veterinary clinic, nutritional status of the animal in the respective study area. It may also be due to the variation in sampling time, as seasonality affects the occurrence of the parasites.

In this study, relatively higher overall prevalence of GIT nematode parasites was recorded in donkey (86.5 %) than horses (55.7%).This study agrees with the study of Tesfu *et al*. (2014)**,** Regassa and Yimer (2013) and Seri *et al*. (2004) in Sudan with their respective results of 78.54%, 70.8% and 37.48% .This current prevalence in donkeys (86.5 %) is higher than the work reported by Gizachew and Ayana (2010) with overall results of 33% *of Parascaris* and *Strongyle* in donkeys in Central Shewa. Horse being infected with prevalence 55.7% of gastrointestinal nematodes agrees with the results reported by Worku and Afera (2012) with overall prevalence of 52% of gastrointestinal nematodes in horses in Kombolcha town. But, it is lower than the result (63.7 %) shown by Tesfu *et al*. (2014) in Hawasa town.

This difference of high prevalence in donkeys might be due to the differences in feeding and deworming activities. Horses in this area are mostly used for cart pulling and they feed on grains and dewormed regularly than donkeys around the town that get less attention. The difference in prevalence in different area might be due to the difference in sampling area, feeding system and accessibility to deworming and health services.

The risk factors, species, sex, age, body conditions and origins with their respective p-value of (0.000, 0.037, 0.028, 0.009 and 0.000), are significantly associated with the infections of GIT nematode parasites. This agrees with the study of Tesfu *et al.* (2014) having significance results for species and age with respective p-value (P=0.001, P=0.043).This also agrees with the study (p=0.000) of Worku and Afera (2012) for both age and body conditions on the study of GIT nematode parasites of horses in Kombolcha town and with the study of Mezgebu *et al*.(2013) for species (P< 0.05) in Gondar.

This current study disagrees with the study of Hailu *et al.* (2013) and Mezgebu *et al*. (2013) who studied respectively in Arsi-Bale highlands of Oromiya Region and in and around Gondar Town. Both showed none significant results for age, sex and body conditions under the study of GIT helminth parasites in equine having p > 0.05.This result also disagrees with the study of Netsanet *et al.* (2014) with his study of none significant p-value (0.362, 0.726), respectively for sex and body conditions. This difference might be due to the difference in the study area, variation in sampling time as seasonality affects the occurrence of the parasites, nutritional status of the animal in the respective study area which can influence the level of immunity to be infected by the parasite, deworming strategy of equines and accessibility to veterinary clinic.

The relative percentage of GIT Nematode parasites reported in this current study indicated that *strongyle* was observed to have higher occurrence rate (43.8 %) than other GIT Nematode parasites. This is in line agreement with the works of Worku and Afera (2012) in Kombolcha town,Wosu and Udob (2014) in Nigeria, Regassa and Yimer (2013) in South wollo zone and Tesfu *et al*.(2014) in Hawassa Town. The 43.8% prevalence of *Strongyles* in donkeys and horses in the current study is higher than the result (32.6%) shown by Worku and Afera (2012) for horses in Kombolcha town. This prevalence is found to be lower than the result (75.26%) of Tesfu *et al* (2014) in Hawasa town, 100% prevalence at the highlands of Wollo Province (Mulate, 2005), Wonchi area (Yosef, 2001) and Western highland of Oromia (Regasa *et al*., 2005). This current higher prevalence for *Strongyles* followed by mixed infections of Strongyle+*Ascaris*, also agrees with the result of Tesfu *et al* (2014).

This difference might be due to the prevalence done for this study for *strongyle* included its single occurrence alone. But it's much proportion found mixedly with other parasites *Ascaris* and *Oxyuris*. Higher infections of *strongyles* correspond with the biology and epidemiology of

these parasites as they require longer period to complete the life cycle and significant change in worm population and their burden under different anthelmintic pressures over the years (Worku and Afera, 2012).

The risk of being infected by GIT nematodes are found high in males (79.7%) than females (70.2%).This was indicated similarly by Regassa and Yimer (2013) and Mezgebu *et al.* (2013). *Strongyles* (44.5%), *P.equorum*(5.9%) and *Strongyle + Oxyuris equi* (6.8%) have their highest prevalence in males while *O. equi* (3.4%) and *Strongyle + P. equarum* (13.5%) are highest in females. This agrees with the studies of Regassa and Yimer (2013). Highest prevalence in males might be due to cart horses in the towns which are all males graze following rivers where the area is moist and cold which create favorable conditions for the growth of the parasites.

The percentage prevalence of Strongyle species was also higher in younger ages. This result also agrees with the study shown by Tesfu *et al* (2014). But, disagree with the results of Regassa and Yimer (2013), which appears higher in olders. This current study indicated GIT nematodes are highly prevalent in youngers (85.1%) followed by older ages (80%). This agrees with the study of Regassa and Yimer (2013) having prevalence of 91.1% and 77.8 % respectively for young and old ages. Higher infection rates and more severe infections indicate a lack of immunity in younger population (Urquhart *et al*., 2003).

P. equarum held high prevalence in poor body conditions (6 / 6.4% /) followed by medium (12 / 5.2% /) and good body mass (3 /5.1% /) while *Strongyles* showed highest prevalence in medium body conditions (117/ 50.6% /) followed by poor (35 / 37.2% /) and good BCS (16 / 27.1% /).These are in line with the study of Worku and Afera (2012).

6. CONCLUSIONS AND RECOMMENDATIONS

This current study indicated high gastrointestinal nematode parasites prevalence in and around Kombolcha town. The major findings from the study were *strongyles, Parascaris equorum*, *Oxyuris equi* and mixed infections of these parasites. Donkeys were at higher risk of infestation than horses. Mostly, the disease affects adults followed by young and old ages. Equines having poor and medium body conditions are highly infected. Age, sex, species, body conditions and origins were the important risk factors for occurrence of gastro intestinal nematode parasites for these species. These risk factors were significantly associated with the occurrence of gastro intestinal nematode parasites. Donkeys in the area was highly infected since they feed by grazing and no care is given for deworming and supplementary feeding, while cart horses are dewormed regularly and feed grains rather than grazing.

Based on the above conclusions, the following recommendations are forwarded:

- Improvement of housing and feeding management system for equines are important.
- Public awareness creation to equine owners on proper regular deworming, sufficient feed supply and minimizing extensive open grazing are also important
- Regular and strategic deworming programmes with efficacious anthelminthics should be carried out regularly.

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8. ANNEXS

Annex 1: Sample collection recording format

		Risk factors					Result		
Id No	Owners name	Animal	Sex	Age(yr)	BCS	Origin	Strongyl	Ascaris	Oxyuris
001	Hassen Ali	Donkey	M	2	Med	Ligo	+	-	-
002	Tasew Belay	Donkey	F	4	Poor	Ligo	+	+	-
003	Ahmed Seid	Hores	M	5	Good	Kombolcha	+	-	-
004	Lema Endalew	Horse	M	3	Medium	Kombolcha	+	-	+

Examples of sample recording format during data collection

Annex 2: Fecal sample flotation procedures

After fecal samples collected from the horse and donkeys, samples were transported to Kombolcha Regional Animal Disease Diagnosis and Laboratory Center and individual sample was processed by the following procedures (Bayou, 2005).

- 3gm of fecal sample was taken and crushed gently using mortar and pestle
- 40ml of flotation fluid were added to the sample in mortar and mixed well.
- The mixture was poured into the plastic beaker using tea strainer.
- This mixture again poured into the test tubes being full to the top.
- The test tube put in upward direction in racks and cover slip added over it
- After 20 minutes of adding the cover slip, the cover slip taken and put over the microscope slide.
- After putting on the microscope slide, the sample examined using 10 and 40 times objective compound microscope

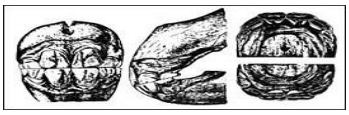
Annex 3: Body condition classifications of donkeys and horses

Body Condition scores	Descriptions
poor	Neck thin and meets shoulder abruptly. Neck and shoulder bones easily felt. Dorsal spine of withers prominent and easily felt. Ribs can be seen from a distance and felt with ease. Belly tucked up. Backbone prominent, dorsal and transverse processes can be felt easily. Hip bones visible and felt easily (hock and pin bones). May be cavity under tail.
Moderate	Some muscle development overlying bones of neck and shoulder. Slight step where neck meets shoulders. Some muscle cover over dorsal withers. Spinous processes of withers felt but not prominent. Ribs not visible but can be felt with ease. Dorsal and transverse processes of back bones felt with light pressure. Poor muscle development on either side of midline. Poor muscle cover on hind quarters. Hip bones felt with ease.
Ideal (Good)	Good muscle/fat development on neck and shoulders, dorsal and transverse spinous processes of withers, ribs, belly, back, hip bones or hindquarters.

Source :(Svendsen, 2008)

Annex 4: Age determinations of donkeys and horses

Under 5 years- This age group has a deciduous UCI from 8 months to 4.5 years. Eruption times of the central, middle, and corner incisors (2.5, 3.5, 4.5 years) are the primary source of information in this age group. One year of age. All temporary teeth are present. The corners are not yet in wear. A typical 3-years-old mouth showing the large permanent center teeth, both upper and lower. Contrast these with the small, light-colored temporary teeth. At 4-years-old, the well-developed permanent centers, immature intermediates and milk teeth at the corners appeared. Tusks or canines have appeared.



5 to 9 years-Age group 5 to 9 has a UCI that progressively changes from wider than tall to square. The upper central incisors should be taller and wider than the middle incisors when viewed from the labial surface. The incisor profile angle should be near 180°.

10 to 14 years -Age group 10 to 14 has a UCI that is square to slightly taller than wide in shape. The occlusal surface of the lower central incisor should begin to resemble a triangle in shape, and the incisor profile angle should begin to change toward a more acute angle.

Source :(Martin *et al.*, 1999)

9. DECLARATION

I the under signed, declare that the information presented here in my thesis is my original work, has not been presented for degree in other university and that all sources of materials used for the thesis have been dully acknowledged.

Name: Samuel Engdaw

Signature: _____

Date of submission: 11/6/2015

This thesis has been submitted for the examination with my approval as University of advisor

Name: Dr Ashenafi Assefa (DVM, MSC)

Signature: _____